hormone secretion, luteinizing hormone, follicle stimulating hormone, prolactin and thyroid stimulating hormone, among others. Gonadal cells, such as Leydig cells and follicular cells are employed to supplement testosterone or estrogen levels. Specially designed combinations are useful in hormone replacement therapy in post and perimenopausal women, or in men following decline in endogenous testosterone secretion. Dopamine-producing neurons are used and implanted in a matrix to supplement defective or damaged dopamine cells in the substantia nigra. In some embodiments, stem cells from the recipient or a donor can be mixed with slightly damaged cells, for example pancreatic islet cells, or hepatocytes, and placed in an electroprocessed matrix and later harvested to control the differentiation of the stem cells into a desired cell type. This procedure is performed *in vitro* or *in vivo*. The newly formed differentiated cells are introduced into the patient.

The ability to use electroprocessed materials and matrices to bioengineer tissue or organs creates a wide variety of bioengineered tissue replacement applications. Examples of bioengineered components include, but are not limited to, skeletal muscle, cardiac muscle, nerve guides, brain constructs as a filler for damaged/removed areas of the brain that are lost during accident or disease, a filler for other missing tissues, cartilage scaffoldings, sheets for cosmetic repairs, skin (sheets with cells added to make a skin equivalent), vascular grafts and components thereof, and sheets for topical applications (skin covering but no additional cells, just a patch). In some embodiments, such matrices are combined with drug and substance delivery electroprocessed matrices of the present invention in ways that will improve the function of the implant. For example, antibiotics, anti-inflammatories, local anesthetics or combinations thereof, can be added to the matrix of a bioengineered organ to speed the healing process and reduce discomfort.

One method or preparing implants of the present invention is use of a bioreactor. There are several kinds of commercially available bioreactors, devices designed to provide a low-shear, high nutrient perfusion environment. Until recently, most of the available bioreactors maintained cells in suspension and delivered nutrients and oxygen by sparging, through the use of impellers, or other means of stirring. The RCCS bioreactor (Synthecon) is a rotating wall bioreactor. It consists of a small inner cylinder, the substrate for the electrospinning process, positioned inside a larger outer cylinder. Although the

electrospun or electroaerosol matrix can be fabricated on the inner cylinder, other locations within the bioreactor also can be used for placement of a matrix for seeding. The gap between the inner and outer cylinders serves as the culture vessel space for cells. Culture medium is oxygenated via an external hydrophobic membrane. The low shear environment of the Synthecon RCCS bioreactor promotes cell-cell and cell-extracellular matrix (ECM) interactions without the damage or "washing away" of nutrients that occurs with active stirring or sparging. Typically, the RCCS device is operated at rotation rates of 8 up to 60 RPM, as required to maintain cells in suspension, and at less than 8 RPM (preferably 2-3 RPM) for cultures immobilized along the center shaft of the vessel. The Synthecon bioreactor can be used in a standard tissue culture incubator. These values for spin rates and other parameters can be varied depending on the specific tissue created.

Electroprocessed materials, such as matrices, are useful in formation of prostheses. One application of the electroprocessed matrices is in the formation of medium and small diameter vascular prostheses. Some preferred materials for this embodiment are collagen and elastin, especially collagen type I and collagen type III. Some examples include, but are not limited to coronary vessels for bypass or graft, femoral artery, popliteal artery, brachial artery, tibial artery, radial artery or corresponding veins. The electroprocessed material is useful especially when combined with endothelial cells on the inside of the vascular prosthesis, and smooth muscle cells, for example a collagen tube, and also when combined with fibroblasts on the outside of the collagen tube. More complicated shapes including tapered and/or branched vessels can also be constructed. A different-shaped mandrel is necessary to wind the large fibers around or to orient the electrospun/electroaerosol polymer.

Combination of electroprocessed matrix materials and wound polymer fibers can provide optimal growth conditions for cells. The polymer forms a basic structural matrix and the electroprocessed matrix is used to deliver the cells. This facilitates cell attachment onto the structural matrix. Furthermore the stress in the polymer also orients fibers in the matrix providing further spatial cues for the cells.

In an alternative fabrication strategy, a cylindrical construct is electrospun onto a suitable target, for example a cylindrical mandrel. Other shapes can be used if desirable based upon the shape of the site into which the implant will be

placed. Matrices in this embodiment are composed, for example, of electroprocessed fibrinogen/fibrin (for example to promote neovascularization, cellular integration and infiltration from the surrounding tissue), electroprocessed collagen (to promote cell infiltration and lend mechanical integrity), and other components, for example PGA, PLA, and PGA-PLA blends, PEO, PVA or other blends. The relative ratio of the different components of this construct is tailored to specific applications (*e.g.* more fibrin, less collagen for enhanced vascularization in a skin graft). To fabricate a cylindrical muscle the construct is filled with muscle or stem cells or other cell type and the distal ends of the electrospun constructs are sutured or sealed shut. In some embodiments, cells are mixed with various matrix materials to enhance their distribution within the construct. For example, the cells can be mixed with electroprocessed fibrin or collagen prior to insertion into the construct. The objective of this strategy is to provide additional mechanical support to the construct and provide the cells with a three dimensional matrix within the construct to promote growth. This also helps to maintain the cells in an even distribution within the construct. This method can be used to enhance the alignment of the cells within the construct. This filling material can be extruded directly into the cylindrical construct, as the filling is extruded, alignment occurs. Mixing endothelial cells with the other cells inserted into the construct (or other cell types) can be done to accelerate neovascularization. Another method to accomplish this objective is to electrodeposit endothelial cells directly into the electroprocessed collagen-matrix that aids in formation of the cylindrical sheath. The alignment of the fibers within the electroprocessed matrix that comprises the construct are optionally controlled by controlling the relative movement of the target and source solution with respect to one another. Other cell types, such as tendon fibroblasts, are optionally electrospun into or onto the outer surface of the construct to enhance the formation of the outer connective tissue sheath that forms the construct.

In another example a sheet of electroprocessed material is prepared, rolled into a cylinder and inserted into an electroprocessed cylinder. The construct is filled with cells as described above, sutured shut and placed in a bioreactor or directly in situ. By aligning the fibrils of the electrospun sheet of material in parallel with the long axis of the outer cylinder a muscle-like, electroprocessed composition is produced. Cells in contact with the fibrils that are arrayed along the long axis of the sheet spread in parallel with the fibrils of the sheet, forming a